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OBSERVATIONS ON THE GERM CELLS OF HYDRA.

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The germ cells of hydra, which pass through stages comparable to those of higher animals, possess some peculiarities that have not been previously mentioned. Some investigators advocate the specificity of the germ cells, while others claim that the sex cells are the immediate derivatives of the ordinary interstitial cells.

Kleinenberg (4), Hertwig (3), Brauer (1) and others consider the sex cells as interstitial in origin and state that the egg first becomes recognizable after the ovary has begun to develop. Downing (2) claims that there is continuity of germ plasm in the sense of a specific line of germ cells, that the egg (Hydra fusca) is always present even before the interstitial cells begin to form the ovary and that the egg may grow rapidly and take the initiative in its formation. He furthermore believes that the egg is recognizable as such in the adult hydra and in general that in some stage in the embryonic development certain cells are stamped with sex characters, so that they and their progeny form the sex cells distinct throughout the life of the individual.

A careful examination of sections from Hydra sp.? (Brauer) gives pretty conclusive evidence that not only the egg but the sperm as well is interstitial in origin. There can be no question in case of the sperm, as the different stages in development can readily be traced from the interstitial cells to the mature sperm. Furthermore, the progenitors of the spermatozoa have no special characters by which they can be recognized as germ cells. The cells that give rise to the eggs are interstitial in position and can be distinguished in the adult hydra from the ordinary interstitial cells by their large nucleus, nucleolus and abundance of chromatin, even before the growth of the ovary begins, as Downing states. This is especially true during the breeding season. If these sex cells could be distinguished during the budding season as well, it would at least suggest specificity of the germ cells.

In the diœcious form Hydra sp.? (Brauer) the egg cells can-

not be distinguished from the ordinary interstitial cells except during the period of sexual reproduction. Fig. 1, a and b, represents two adjacent eggs at the stage of development when they first become recognizable. They have begun to enlarge and can readily be distinguished from the adjoining cells by their size and vacuoles next the nucleus. The above figure was taken from an adult hydra in which six eggs were present in different stages of development, ranging from the interstitial cells to the undisputed egg. These eggs are isolated or found in groups. When two or more are found adjacent the cell walls become dissolved and one persists as the developing egg (Tannreuther). In a few instances observed two of these adjacent cells persisted and gave rise to two mature eggs. These cases, however, are extremely rare in proportion to the number of eggs produced.

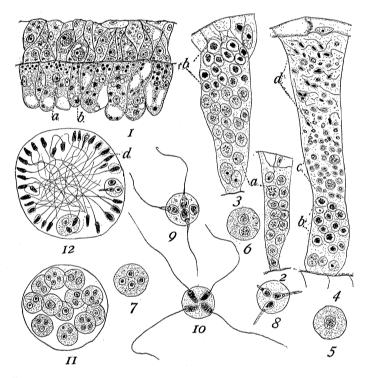
The above results do not warrant the view that there is continuity of germ plasm in Hydra. Until sex cells distinct from the somatic can be traced through successive generations, we have no positive evidence of such a continuity.

The different generations of the sex cells of *Hydra* are distinct and can readily be recognized. In the formation of the egg there is a distinct growth period. Reduction occurs at the end of the growth period just before the first polar body is formed. The polar bodies remain attached after cleavage has begun, by means of a single cytoplasmic thread. The first polar body is larger than the second.

The spermaries are composed of an indefinite number of cysts. The individual cysts originate from a single or several interstitial cells. Fig. 2 represents a longitudinal section of a single cyst containing spermatogonia, which have originated from a single interstitial cell. After the spermatogonia have divided a few times (the number of divisions varies in different individuals) those found at the distal end of the cyst become transformed into the spermatocytes of the first generation without growth. In Fig. 3 the spermatocytes of the first generation are recognized by their dense chromatin mass. Different stages of development are found in a single cyst, ranging from the spermatocytes of the first generation to mature sperm, as shown in Fig. 4. The successive zones of the developing sperm are distinct. The nuclei

at the extreme proximal end of the cyst cease dividing and become the nuclei of new interstitial cells after the spermaries have disappeared.

My observations on the nuclear changes in the different stages of development confirm those of previous investigators. Reduction occurs after the last spermatogonial division just before the spermatocytes of the first generation divide. In the division of the nuclei of the spermatocytes of the first and second generations, the cell wall remains intact and produces a multinucleate cell (Figs. 5-7), and give rise to four sperm within a common



- Fig. 1. Longitudinal section showing two developing eggs. a and b, eggs.
- Fig. 2. Longitudinal section of a single cyst.
- Fig. 3. Section of cyst little later than preceding. a, spermatogonia; b, spermatocytes of the first generation.
- Fig. 4. Longitudinal section of cyst showing maximum development.
- c, spermatocytes of the first and second generations; d, mature sperm.
 - Figs. 5-7. Formation of spermatids within common vesicle.
 - Figs. 8-10. Developing sperm. Figs. 5-10, from living cells.
 - Figs. 11 AND 12. Cysts removed from living spermary.

vesicle. The vesicular wall is very thin and the four sperm pass through it when mature and become free within the distal end of the cyst (Fig. 4). The mature sperm are very active within the cyst and finally escape to the exterior through a small temporary opening of the spermary. After the mature sperm have all escaped, the opening closes until more sperm mature. When the spermaries are extremely large, this process continues from forty to fifty hours.

In order to show the four sperm within a common vesicle, it is necessary to dissect the living spermary apart, as the sperm always escape from the common vesicle before passing to the exterior. The individual cysts when separated become more spherical (Figs. 11 and 12), and the different stage of development in the living sperm can easily be distinguished.

Kleinenberg in his description of the sperm in Hydra viridis definitely states that the sperm are formed from interstitial cells that have divided a number of times; ultimately the nucleus of the cell (the spermatocyte of the first order) disintegrates while the cell substance becomes granular, and in place of the nucleus there appears from one to four refractive bolies, which give rise to the sperm. The refractive bodies referred to beyond doubt result from the two divisions of the nucleus, which he thought disintegrated. The four nuclei (spermatids) within the common vesicle do have the appearance of refractive bodies, especially in the living material.

Korotneff (5) gave similar results. He states that the sperm form directly from the nuclei of a multinucleate mother cell. Downing (2) does not mention this interesting phenomenon, which is found in *Hydra viridis* and *Hydra* sp.? (Brauer).

The mature sperm possess extreme vitality and may remain active from one to three days after escaping from the spermary. The mature sperm of any individual spermary possess about the same degree of fertility. No degenerating spermatogonia were found.

In the monœcious form *Hydra viridis* the spermaries become mature before the ovaries. Occasionally sperm and ova on one individual would ripen at the same time, making self-fertilization possible. In order to prove that self-fertilization did occur, indi-

viduals with both spermaries and ovaries at the same stage of development were isolated and placed in distilled water for one hour, in order to kill any mature sperm that might adhere from other individuals. The *Hydra* were then placed separately in water free from sperm. The spermaries and ovaries matured and self-fertilization took place. The cleavage was normal.

SUMMARY.

- I. Hydra sp.? (Brauer) does not show continuity of the germ plasm.
- 2. The eggs can be distinguished from the interstitial cells in the adult *Hydra* before the ovary is formed. This is especially true during the breeding season.
- 3. The different generations in the formation of the germ cells are comparable to those of higher animals.
- 4. In the division of the spermatocytes of the first and second generations the cell wall remains intact and the four spermatids are formed within a common vesicle, each producing a mature sperm.
 - 5. Self-fertilization occurs in Hydra viridis.

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